

to compare it with the pollen of *Nothoscordum borbonicum* Kunth, *Tulbaghia simmleri* P. Beauv. en *T. violaceae* Harv. Floral material of the four species was collected from the gardens of the North-West University (NWU) campus and a special collection held at the NWU-botanical garden. Pollen was examined with scanning electron- and light microscopy. The pollen morphology of the four species is perprolate and monosulcate, and the surface sculpturing is reticulate and heterobrochate. The pollen of *Prototulbaghia siebertii* however, displays a unique characteristic as the grains fold breadth-wise causing the tips to touch. Hence, the grain displays a triangular shape and has a disulcate appearance. Two hypotheses can be formulated to explain this phenomenon. The folding can either be due to the process of harmomegathy or a still unknown event that occurs during the development of the pollen grain. These processes will be investigated further to determine the mechanism of the folding, whether it is a unique taxonomic character for the genus, and whether it is of evolutionary significance within the Alliaceae.

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### Regulation of bioactive secondary metabolite production in *in vitro*-derived greenhouse-grown *Aloe arborescens*

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Auxins and cytokinins are known to interact at different levels in synergistic, antagonistic or additive ways to produce or regulate physiological effects. The effects of cytokinins as well as auxin and smoke-water (SW) treatments applied during the shoot regeneration and rooting phases respectively, on secondary metabolite production of *in vitro*-derived *Aloe arborescens* were evaluated after two months of growth in the greenhouse. The cytokinins used during shoot regeneration were 6-benzyladenine (BA), *meta*-topolin (*mT*) and their derivatives while indole-butyric acid (IBA) and SW were used for rooting. In *MemT*-regenerated shoots, the treatment with either SW or IBA during rooting significantly increased total phenolic and flavonoid contents when compared to those rooted without plant growth regulators (PGR). On the other hand, *mT*, *MemTR* and *MemTTHP* regenerated shoots which were rooted on PGR-free medium, yielded a significantly higher flavonoid content when compared to those rooted on SW and/or IBA-containing medium. Regenerated shoots from BAR-containing medium rooted using SW treatment gave a significantly higher iridoid level compared to those rooted using IBA or without PGR. Conversely, significantly lower iridoid levels were observed with regenerated shoots from BA, *mT* and *mTR*-containing media rooted with IBA or SW when compared to those rooted without PGR. Regenerated shoots from *mTR* and *MemT*-containing media rooted with SW showed a significantly reduced radical scavenging activity when compared to those rooted without PGR. Shoots regenerated from BA, BAR and *MemTTHP*-containing media and rooted with SW demonstrated a significantly higher radical scavenging activity when compared to those rooted without PGR. Taken together, the cytokinin type used during shoot proliferation stages and the rooting treatment applied, individually and interactively had a significant carry-over effect on the production of bioactive secondary metabolite in *A. arborescens*.

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### The role of cytokinins on the antioxidant capacity and phenolic acid content during tissue culture and acclimatization of *Merwillia plumbea*

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*Merwillia plumbea* (Lindl.) Speta, a member of the Hyacinthaceae is a popular South African medicinal plant which is currently threatened in the wild. Cultivation using tissue culture techniques could alleviate pressure on wild populations. Nevertheless, there is no adequate literature on the phytochemical and pharmacological efficacy of tissue cultured *M. plumbea*. In addition effects of the series of events that occur during tissue culture and acclimatization on the production of bioactive constituents remain intricate and poorly-understood. Hence, we evaluated the effect of five cytokinins (isoprenoid and aromatic) on the antioxidant activity and phenolic acid content of *M. plumbea* during tissue culture and acclimatization stages. *Meta*-topolin riboside-treated and control plantlets had the best ORAC activity during tissue culture and acclimatization stages, respectively. Generally, the antioxidant activity increased in the aerial parts and decrease in the underground parts after acclimatization. Remarkably, the level of phenolic acids such as proto-catechuic acid, 4-hydroxybenzoic acid, caffeic acid and vanillic acid were generally higher in the plant parts and stages which demonstrated better antioxidant activity. The current findings highlight the great chemical variations that could result from the type of applied cytokinin. The use of the right cytokinin is required to guarantee the presence of bioactive chemicals and subsequently the efficacy of micropropagated *M. plumbea*.

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### Are long-term stored medicinal plants as active as the freshly harvested materials?

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Medicinal plant materials are usually stored for a period of time by plant gatherers, traders or traditional healers before usage. Questions that frequently arise are: Do such plant materials retain their biological efficacy?; Are they safe for consumption after a long period of storage? In the present study, the iridoid and phenolic contents as well as the antioxidant and mutagenic properties of five South African medicinal plants stored for 16 years were compared to those of freshly harvested materials. The iridoid content of the freshly harvested materials of *Ocotea bullata*, *Protorhus longifolia* and *Ziziphus mucronata* were significantly higher when compared to the stored materials. Stored *Acokanthera oppositifolia* material had a significant higher iridoid content compared to the freshly harvested material. The total phenolic and flavonoid contents recorded in all freshly collected materials (except *Artemisia afra*) were significantly higher than the